REF 130219026M:100 tests
130619026M: 50 tests

MAGLUMI® SARS-CoV-2 Ag (CLIA)

INTENDED USE
The kit is a chemiluminescence immunoassay for the quantitative determination of SARS-CoV-2 nucleocapsid protein in nasopharyngeal swab and oropharyngeal swab specimens using the MAGLUMI series Fully-auto chemiluminescence immunoassay analyzer.

SUMMARY AND EXPLANATION OF THE TEST
The Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) causes an epidemic of acute respiratory syndrome in humans[1]. SARS-CoV-2 is a single-stranded RNA virus, it has an envelope, particles are round or oval, often polymorphic, and the diameter is 60 ~ 140nm. SARS-CoV-2 encodes four structural proteins: spike, envelope, matrix, and nucleocapsid [2]. Genetic sequencing of the virus suggests that it is a betacoronavirus closely linked to the SARS virus [3]. SARS-CoV-2 is mainly transmitted through respiratory droplets and can also be transmitted through contact. The sources of infection seen so far are mainly patients with pneumonia infected by the novel coronavirus.
The SARS-CoV-2 is present in the upper respiratory tract, such as the nose and throat. Standard confirmation of acute SARS-CoV-2 infections is based on the detection of unique viral sequences by nucleic acid amplification tests (NAATs), such as real-time reverse-transcription polymerase chain reaction (rRT-PCR). The assays' targets include regions on the E, RdRP, N and S genes [2]. Antigen is generally detectable in nasal swabs during the acute phase of SARS-CoV-2 infection. Antigen-detection diagnostic tests are designed to directly detect SARS-CoV-2 proteins produced by replicating virus in respiratory secretions and have been developed as both laboratory-based tests, and for near-patient use, socalled rapid diagnostic tests, or RDTs [4].

PRINCIPLE OF THE TEST
The SARS-CoV-2 Ag (CLIA) assay is a sandwich chemiluminescence immunoassay.
The swab sample (treated by Buffer), magnetic microbeads coated with anti-nucleocapsid protein monoclonal antibody, ABEI labeled with another anti-nucleocapsid protein monoclonal antibody are mixed thoroughly, reacting to form sandwich complexes and incubating. After precipitation in a magnetic field, the supernatant is decanted and then a wash cycle is performed. Subsequently, the Starter 1+2 are added to initiate a chemiluminescent reaction. The light signal is measured by a photomultiplier as relative light units (RLUs), which is proportional to the concentration of nucleocapsid protein present in the sample.

KIT COMPONENTS
Material Provided

Component	Contents	100 tests	50 tests
Magnetic Microbeads	Magnetic microbeads coated with anti-nucleocapsid protein monoclonal antibody (mouse) in PBS buffer with BSA, NaN_3 (0.09%).	2.5 mL	1.5 mL
Calibrator Low	A low concentration of nucleocapsid protein antigen in PBS buffer with BSA, NaN_3 (0.09%).	2.0 mL	1.5 mL
Calibrator High	A high concentration of nucleocapsid protein antigen in PBS buffer with BSA, NaN_3 (0.09%).	2.0 mL	1.5 mL
Buffer	Tris-HCl buffer with surfactants, NaN_3 (0.09%).	6.5 mL	4.0 mL
ABEI Label	ABEI labeled with anti-nucleocapsid protein monoclonal antibody (mouse) in PBS buffer with BSA, NaN_3 (0.09%).	7.5 mL	4.5 mL
Control 1	A low concentration of nucleocapsid protein antigen in PBS buffer with BSA, NaN_3 (0.09%).	2.0 mL	1.5 mL
Control 2	A high concentration of nucleocapsid protein antigen in PBS buffer with BSA, NaN_3 (0.09%).	2.0 mL	1.5 mL
All reagents are provided ready-to-use.			

Required But Not Provided

MAGLUMI Series:	Reaction Module	REF: 630003
	Starter 1+2	REF: 130299004M; 130299027M
	Wash Concentrate	REF: 130299005M
	Light Check	REF: 130299006M
	Reaction Cup	REF: 130105000101
	Maglumi 600	REF: 23020018
	Maglumi 800	REF: 23020003
	Maglumi 1000	REF: 23020009
	Maglumi 2000	REF: 23020006
	Maglumi 2000 Plus	REF: 23020007
	Maglumi 4000	REF: 23020014
	Maglumi 4000 Plus	REF: 23020037
	MAGLUMI X8	REF: 010101008801
Other Material	Sample Extraction Solution	REF: 130299030M, 130299031M, 130299032M, 130299033M
	Nylon flocked nasopharyngeal swab, nylon flocked oropharyngeal swab	
	Universal Transport Media (UTM), Viral Transport Media (VTM)	

Please order all above from Shenzhen New Industries Biomedical Engineering Co., Ltd. (SNIBE) or our authorized representative.

CALIBRATION
Traceability: This method has been standardized against the SNIBE internal reference substance.
Test of assay specific calibrators allows the RLU values to adjust the assigned master curve. Results are determined via a calibration curve which is instrument-specifically generated by 2-point calibration and a master curve provided via the reagent Radio Frequency Identification (RFID) CHIP.
Recalibration is recommended if any of the following conditions occurs:
- After each exchange of lots (Reagent or Starter 1+2).
- Every 2 weeks and/or each time a new reagent kit is used.
- After instrument service is required.
- If controls lie outside the expected range.

QUALITY CONTROL

Follow local regulations or accreditation requirements for quality control frequency.
The controls in kit are only applicable with MAGLUMI system and used matching with the same LOT numbers of corresponding reagents. For each target value and range refer to the label.
To monitor system performance, quality control materials (negative control and positive control) are required. Treat all quality control samples with the same level of care as patient samples. A satisfactory level of performance is achieved when analyte values obtained are within the acceptable Control Range for the system or within your range, as determined by an appropriate internal laboratory quality control scheme. If the quality control results do not fall within the Expected Range or within the laboratory's established Range, measurement of the quality control should be repeated. If the quality control results still do not fall within the range, do not report results and take the following actions:

- Verify that the materials are not expired.
- Verify that required maintenance was performed.
- Verify that the assay was performed according to the instruction for use.
- Rerun the assay with fresh quality control samples.
- If necessary, contact your local technical support provider or distributor for assistance.

SPECIMEN COLLECTION AND PREPARATION

Specimen Types

- Nasopharyngeal swab and oropharyngeal swab transported in Universal Transport Media (Copan, UTM) or Viral Transport Media (MANTACC, VTM) *, then processed following the pre-analytical procedure indicated below.
- Dry nasopharyngeal swab and oropharyngeal swab processed following the below indicated pre-analytical procedure.

* UTM and VTM from other manufacturers have not yet been verified.

WARNING: for the collection and handling of swab specimens from the upper respiratory tract, refer to the local regulations covering the collection, handling and transportation of clinical specimens from persons with COVID-19, and to the WHO's Interim guidance of Diagnostic testing for SARS-CoV-2 at https://www.who.int/publications/i/item/diagnostic-testing-for-sars-cov-2. Refer to the WHO website for additional publications.

Pre-analytical Procedure Workflow for Swab in UTM or VTM

- Collect the sample by nasopharyngeal swab and oropharyngeal swab transported in UTM/VTM.
- All clinical samples must be at room temperature (15-25°C) before beginning the next step of the procedure.
- Pipette at least 0.6 mL of the specimen eluted in UTM/VTM into the sample container (such as 1.5 mL centrifuge tube). To obtain accurate results, avoid mucoid substances when pipetting sample in UTM/VTM.
- Cap the tube and centrifuge at 1000×g for 5 minutes. Load the sample into the instrument for testing.

Pre-analytical Procedure Workflow for Dry Swab -Use Sample Extraction Solution (REF: 130299030M, 130299031M, 130299032M)

- Collect nasopharyngeal swab and oropharyngeal swab samples according to clinical laboratory standard methods.
- Open the purple cover of the pre-filled tube containing the Sample Extraction Solution (REF: 130299030M, 130299031M, 130299032M), place and soak the nasopharyngeal swab and oropharyngeal swab in liquid.
- Squeeze the lower end of the tube, roll the swab at least 3 times and remove the swab up and down 3 times. Be careful not to spill the liquid.
- Leave the swab in the tube at room temperature (15-25°C) for 5-6 minutes.
- Remove the swab while squeezing the sides of the tube to extract the liquid from the swab, and tightly sealed the tube with the purple cap. Dispose of the used swab in the biohazardous waste collection.
- Transport to the testing instrument, gently invert the tube 5 times. Open the top white cover of the tube, squeeze the middle of the tube, transfer the liquid to the sample container (such as 1.5 mL centrifuge tube) by dripping, and load the sample into the instrument for testing.

Pre-analytical Procedure Workflow for Dry Swab -Use Sample Extraction Solution (REF: 130299033M)

- Collect nasopharyngeal swab and oropharyngeal swab samples according to clinical laboratory standard methods.
- Prepare a 1.5mL/2.0mL centrifuge tube, pipette 1.0 mL Sample Extraction Solution (REF: 130299033M) into the tube, place and soak the nasopharyngeal swab and oropharyngeal swab in liquid.
- Rotate the swab in the liquid at least 5 times, and remove the swab up and down 5 times. Be careful not to spill the liquid.
- Leave the swab in the tube at room temperature (15-25°C) for 5-6 minutes.
- Take out the swab and close the centrifuge tube cap. Centrifuge at 1000×g for 5 minutes. Load the sample into the instrument for testing.

Samples should be tested as soon as possible after the completion of the pre-analytical procedure. If immediate testing is not possible, samples can be stored at 2-8°C for up to 7 days or at -70°C for long-term storage. If samples are stored frozen, mix thawed samples well before testing. Frozen samples can undergo up to 4 freeze/thaw cycles.

Please pay attention to the risk of infection during sample collection and preparation.

The sample volume required for a single determination is 150 µL.

WARNING AND PRECAUTIONS FOR USERS

- *For Research Use Only.*
- Follow the package insert carefully. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this package insert.
- For best performance, samples should be tested as soon as possible after collection.

Safety Precautions

- **CAUTION:** This product requires the handling of human specimens. Biosafety Level 2 or other appropriate biosafety practices should be used for materials that contain or are suspected of containing infectious agents.
- All samples, biological reagents and materials used in the assay should be considered potentially able to transmit infectious agents. They should therefore be disposed of in accordance with the practices of your institution. Discard all materials in a safe and acceptable manner and in compliance with prevailing regulatory requirements.
- This product contains Sodium Azide. Dispose of contents and container must be in accordance with all local, regional and national regulations.
- Refer to safety data sheets, which are available on request.

Handling Precautions

- Do not use reagent kits beyond the expiration date.
- Do not interchange reagent components from different reagents or lots.
- Prior to loading the Reagent Kit on the system for the first time, the Reagent Kit requires mixing to re-suspend magnetic microbeads that have settled during shipment. For magnetic microbeads mixing instructions, refer to the Preparation of the Reagent section of this package insert.
- To avoid contamination, wear clean gloves when operating with a reagent kit and sample.
- Over time, residual liquids may dry on the septum surface. These are typically dried salts which have no effect on assay efficacy.
- To avoid evaporation of the liquid in the opened reagent kits in refrigerator, it is recommended that the opened reagent kits to be sealed with reagent seals contained within the packaging. The reagent seals are "single use", and if more seals are needed, please contact SNIBE or our

authorized representative.
- For detailed discussion of handling precautions during system operation, refer to the SNIBE service information.

STORAGE AND STABILITY
- Store at 2-8°C. Do not freeze.
- Keep upright for storage to facilitate later proper resuspension of magnetic microbeads.
- Keep away from sunlight.
- The stability study is still on-going, the following data is obtained by referring to similar products:

Stability of the reagent	
unopened at 2-8°C	until the stated expiration date
opened at 2-8°C	6 weeks
onboard	4 weeks

- To ensure the best kit performance, it is recommended to place opened kits in the refrigerator after the end of the intraday test work.
- The controls in kit can be stable until the expiration date at 2-8°C in an unopened state, and can be stable for 6 weeks when properly stored at 2-8°C after opening, or stored up to 2 months frozen at -20°C or colder. Frozen and thawed for 2 times.

TEST PROCEDURE
Preparation of the Reagent
- Take the reagent kit out of the box and observe the sealing film and other parts of the reagent kit to see if there is any leakage. In case of leakage, please contact your local distributor immediately. And then tear off the kit sealing film carefully.
- Open the reagent area door; Hold the reagent handle to get the RFID label close to the RFID reader (for about 2s); the buzzer will beep; one beep sound indicates successful sensing.
- Keeping the reagent straight insert to the bottom along the blank reagent track.
- Observe whether the reagent information is displayed successfully in the user software interface, otherwise repeat the above steps.
- Resuspension of the magnetic microbeads takes place automatically when the kit is loaded successfully, ensuring the magnetic microbeads are totally resuspended homogenous prior to use.

Assay Calibration
- Click **<Calibration>** or **<Batch Calibration>** button to execute calibration operation. For specific information on ordering calibrations, refer to the Calibration Section of the Operating Instructions.
- Execute recalibration according to the calibration interval required in this package insert.

Quality Control
- In order to avoid manually error in entry of control information, the provided barcode labels of quality control in the kit can be used attached on the test tubes.
- If users do not use the provided barcode labels for positive and negative controls contained within the packaging, then quality controls should be ordered manually.
- The controls in kit must be edited as “CONTROL” in MAGLUMI series Fully-auto chemiluminescence immunoassay analyzer user software. For specific information on ordering quality controls, refer to the Quality Control Section of the Operating Instructions.

Sample Testing
- Order the samples in the Sample Area of the software and click the **<Start>** button to execute testing. For specific information on ordering patient specimens, refer to the Sample Ordering Section of the Operating Instructions.
- To ensure proper test performance, strictly adhere to the operating instructions of MAGLUMI series Fully-auto chemiluminescence immunoassay analyzer.

LIMITATIONS
- The kit does not differentiate between SARS-CoV and SARS-CoV-2.
- This test is suitable only for investigating single samples, not for pooled samples.
- Bacterial contamination may affect the test results.
- Assay results should be utilized in conjunction with patient history and other clinical and laboratory methods information to assist the clinician in making individual patient diagnostic decisions.
- Assay results should not be used as the sole basis for the diagnosis and exclusion of COVID-19, but only as a supplement to existing viral nucleic acid detection reagents and imaging features.
- If the SARS-CoV-2 Ag results are inconsistent with clinical evidence, additional testing is suggested to confirm the result.

RESULTS
Calculation of Results
The analyzer automatically calculates the concentration in each sample by means of a calibration curve which is generated by a 2-point calibration master curve procedure. The results are expressed in pg/mL. For further information please refer to the operating instructions of MAGLUMI series Fully-auto chemiluminescence immunoassay analyzer.

Interpretation of Results
Results of reference range study using the SARS-CoV-2 Ag assay was as follows:
- Non-reactive: A result less than 25.0 pg/mL (<25.0 pg/mL) is considered to be non-reactive.
- Reactive: A result greater than or equal to 25.0 pg/mL (≥25.0 pg/mL) is considered to be reactive.
- For samples with concentration near the cut-off or positive, follow-up tests should be performed.
- Results may differ between laboratories due to variations in population. It is recommended that each laboratory establish its own expected ranges.

PERFORMANCE CHARACTERISTICS
Precision
Precision for SARS-CoV-2 Ag assay was determined as described in the CLSI EP05-A3. 2 controls and 3 human oropharyngeal swab samples containing different concentration of analyte were assayed in duplicate at three sites on five days, with 3 runs per day, one lot of reagent for each run and 2 replicates per run. The result is summarized in the following table:

Sample	Mean Value (pg/mL)	N	Repeatability		Between-Lot		Between-Day		Between-Site		Reproducibility	
			SD (pg/mL)	%CV	SD (pg/mL)	%CV	SD (pg/mL)	%CV	SD (pg/mL)	%CV	SD (pg/mL)	%CV
QC1	139.589	90	2.861	2.05	0.517	0.37	0.736	0.53	0.611	0.44	3.060	2.19
QC2	718.614	90	8.637	1.20	3.674	0.51	2.262	0.31	0.289	0.04	9.659	1.34
S1	99.701	90	2.244	2.25	1.150	1.15	0.787	0.79	0.497	0.50	2.688	2.70
S2	298.594	90	6.618	2.22	1.271	0.43	0.243	0.08	0.578	0.19	6.768	2.27
S3	1198.15	90	21.584	1.80	2.584	0.22	6.817	0.57	1.634	0.14	22.841	1.91

Limit of Blank (LoB)
The LoB for SARS-CoV-2 Ag assay is 4.00 pg/mL.

Limit of Detection (LoD)
The LoD for SARS-CoV-2 Ag assay is 8.00 pg/mL.

Limit of Quantitation (LoQ)

The LoQ for SARS-CoV-2 Ag assay is 12.0 pg/mL.

Linear Range
12.0 - 10000 pg/mL (defined by the Limit of Quantitation and the maximum of the master curve).

High-Dose Hook
No high-dose hook effect was seen for SARS-CoV-2 Ag concentrations up to 100,000 pg/mL.

Interference
The following substances, naturally present in respiratory specimens or that may be artificially introduced into the nasal cavity or nasopharynx, were evaluated with the SARS-CoV-2 Ag assay at the concentrations listed below and were found not to affect test performance.

Substance	Active Ingredient	No interference up to	Substance	Active Ingredient	No interference up to
Endogenous	Mucin	2% w/v	Throat Lozenge	Menthol	0.15% w/v
	Whole Blood	1% v/v	Anti-viral Drug	Oseltamivir Phosphate	0.5% w/v
OTC Nasal Drops	Phenylephrine	15% v/v	Nasal Ointment	Mupirocin	0.25% w/v
OTC Nasal Gel	Sodium Chloride	5% v/v	Antibacterial Drug	Fluconazole	5% w/v

Cross-Reactivity
The cross-reactivity study for the SARS-CoV-2 Ag (CLIA) assay was designed to evaluate potential cross reactants that may be present in the nasal cavity. The results are listed in the following table:

Category	N of samples	Reactive	Category	N of samples	Reactive
Human Coronavirus (OC43, NL63, 229E)	8	0	Adenovirus	8	0
Influenza A virus	5	0	Mycoplasma pneumoniae	10	0
Influenza B virus	7	0	Chlamydia pneumoniae	12	0
Respiratory syncytial virus	6	0	Legionella pnuemophila	8	0
MERS coronavirus	4	0	Streptococcus pneumoniae	10	0
Parainfluenza virus（1,2,3,4)	6	0	Bordetella pertussis	4	0
Rhinovirus	7	0	Staphylococcus aureus	4	0

Clinical Sensitivity
The clinical sensitivity of the SARS-CoV-2 Ag assay was determined by testing 121 samples confirmed COVID-19 infected specimens by RT-PCR.

Days Post Onset of Symptoms	N of samples	Reactive	Sensitivity	95% CI
0-7	121	118	97.5%	95.2%-100.0%

The positive rate of SARS-CoV-2 Ag may be affected by the infection period of the test subject in different studies.

Clinical Specificity
The clinical specificity of the SARS-CoV-2 Ag assay was determined by testing 236 samples from subjects assumed to be negative for SARS-CoV-2.

N of samples	Non-reactive	Specificity	95% CI
236	235	99.6%	97.8%-100.0%

REFERENCES

1. WHO. Laboratory testing for coronavirus disease (COVID-19) in suspected human cases. Interim guidance 19 March 2020.
2. WHO. Diagnostic testing for SARS-CoV-2. Interim guidance 11 September 2020.
3. WHO. Clinical management of COVID-19. Interim guidance 27 May 2020.
4. WHO. Antigen-detection in the diagnosis of SARS-CoV-2 infection using rapid immunoassays. Interim guidance 11 September 2020.

Shenzhen New Industries Biomedical Engineering Co., Ltd.
No.23, Jinxiu East Road, Pingshan District, 518122 Shenzhen, P.R. China
Tel: +86-755-21536601 Fax: +86-755-28292740

SYMBOLS EXPLANATIONS

	Consult instructions for use		Manufacturer
8 °C 2 °C	Temperature limit (Store at 2-8°C)		Use-by date
Σ	Contains sufficient for <n> tests		Keep away from sunlight
	This way up	CONTENTS	Kit components
REF	Catalogue number	LOT	Batch code